

Abstract

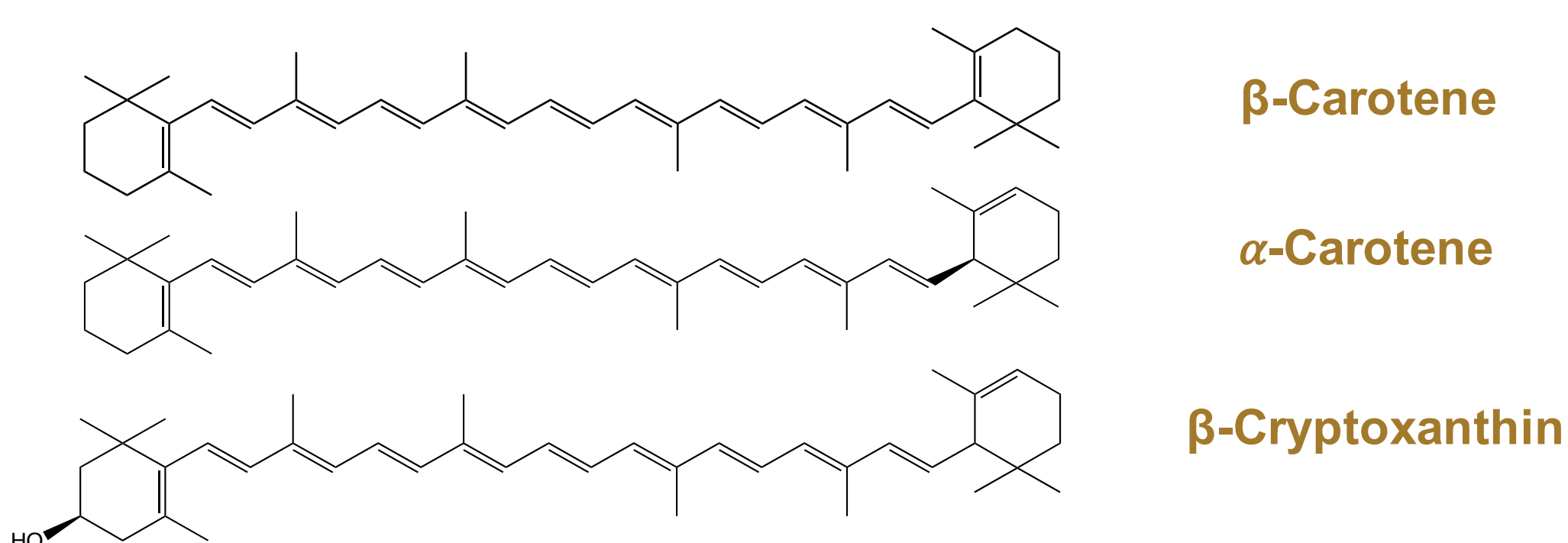
Carotenoid content in cassava roots has been increased through biofortification programs as a strategy to combat vitamin A deficiency. However, incorporation of biofortified cassava into both traditional and industrial food processing has yet to be fully assessed. The objective of this study was to examine the impact of fermentation and thermal processing on the stability and bioaccessibility of pro-vitamin A carotenoids from distinct biofortified cassava roots. Unfermented (UF) and fermented (F) flours were produced from 10 biofortified cassava cultivars (**Table 1 & Figure 2**). Gari (G) flours were produced by toasting two of the fermented cultivars above at 150-160°C during 15-20 min. Test porridges were prepared with UF, F and G (22.2% W/V) in boiling water for 5 min. Bioaccessibility of pro-vitamin A carotenoids was then evaluated from finished products using a three-stage *in vitro* digestion model (**Figure 3**). Overall, cassava cultivars contained 23.1-42.7 µg of β-carotene equivalents (β-CE) / g on dry weight bases (DW). β-CE retention after fermentation was 72.5-96.6%; after oven-drying were 18.3-77.5% and 45.8-80.4% for UF and F roots, respectively; after toasting in Gari preparation was 67.3-69.2%; after cooking in porridge preparations were 42.5-74.5%, 20.7-77.3% and 87.2-115.3% for UF, F and G flours, respectively (**Figure 4 & 5**). Cassava flours, which involved fermentation showed higher β-CE retention (p=0.007) during oven-drying compared with UF flours. However, no significant differences were found in β-CE retention during porridge preparation (p=0.905). Test porridges made from UF, F and G flours ranged from 39-309, 58-343 and 223-323 µg β-CE / 100 g FW, respectively. Bioaccessibility ranged widely from 3.3-56.9 µg β-CE / 100 g FW with bioaccessible content among the cultivars within the porridges groups ranging from 3.3-43.4, 3.66-21.4 and 20.3-56.9 µg / 100 g FW for UF, F and G flours, respectively (**Figure 6**). In general, bioaccessible β-CE content from porridges prepared with UF and F flours were similar with levels of 14.5 ±4.2 and 12.7 ±1.8 µg / 100 g FW, (p = 0.700). Select cassava cultivars showed improved bioaccessibility of β-CE content with the fermentation process, these results suggest that genotype factor and/or another factors in the matrix merit further investigation as they may play a significant role in facilitating bioaccessibility of carotenoids from biofortified cassava products.

Introduction

Vitamin A deficiency (VAD) affects approximately 190 million preschool-age children and 19 million pregnant women globally.¹ VAD is the principal cause of blindness in children, as well as stunting of growth and contributing to morbidity and mortality in these populations. ^{2,3} Biofortification of staple crops is one of the strategies proposed to address VAD along other strategies such as supplementation and fortification. Biofortification is a strategy focused on developing of micronutrient dense staple crops (e.g.: increased pro-vitamin A carotenoids content) that can be leveraged as cost-effective alternative to combat vitamin A deficiency.⁴ This strategy is reliant on the ability to incorporate biofortified staple crops into traditional and industrial food preparations. Insights into acceptability by consumers as well as recovery/bioavailability of pro-vitamin A carotenoids in these food products is lacking.

Cassava (*Manihot esculenta*, Crantz) is a staple crop consumed for more than 70 million people in developing countries and has been included into biofortification programs with the goal of increasing its pro-vitamin A carotenoid content.⁵ Cassava roots are traditionally subjected to both thermal and fermentative processes in order to minimize cyanogen content, extend shelf-life and facilitate marketing of cassava products in urban areas.⁶ However, the impact of such processes on retention of provitamin A in biofortified cassava cultivars is not well understood.

Figure 1. Pro-Vitamin A Carotenoids found in biofortified cassava roots :



Objective

The specific objective of this study was to examine the impact of fermentation and thermal processing on the stability and bioaccessibility of pro-vitamin A carotenoids from a selected group of biofortified cassava roots.

Acknowledgments

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Materials & Methods

Table 1: Cassava root cultivars*

GM 4414-5	GM 4571-3	GM 5194-5	GM 5194-13	GM 5212-6
SM 3757-75	SM 3758-43	SM 3762-15	SM 3767-84	SM 3774-21

*All germoplasms were provided by CIAT.

Figure 2. Cassava root processing⁶

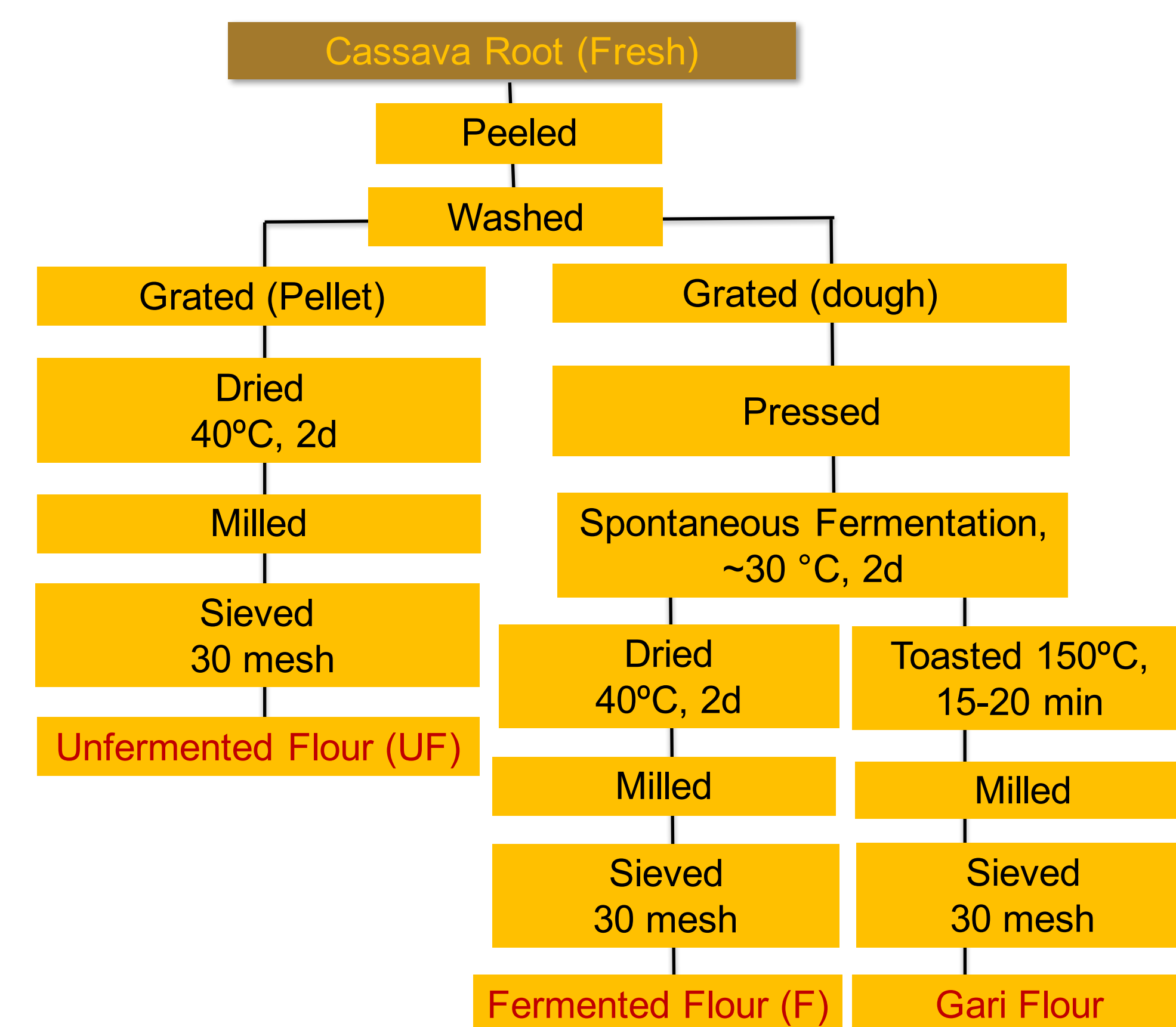
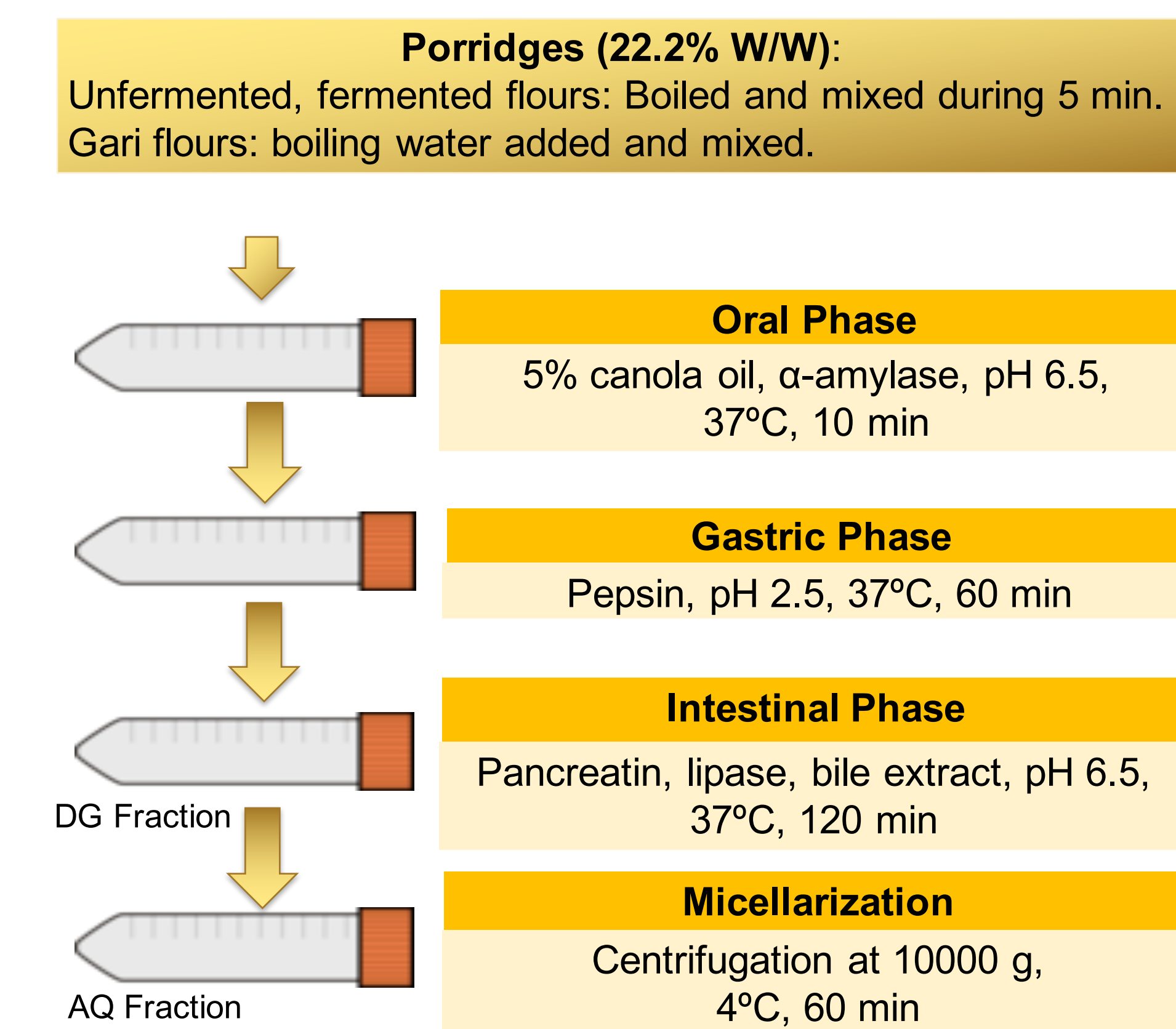


Figure 3. Three stages *in vitro* digestion⁷



Extraction and carotenoids analysis: Extraction⁵ and quantitation⁷ were based on previously reported methods. Briefly, cassava samples were extracted with 10 mL petroleum ether and 10 mL acetone (X3). Then, collected organic extracts were washed with saline solution (0.1 M NaCl), dried under a stream of nitrogen, solubilized in 1:1 methanol:ethyl acetate, and then analyzed by HPLC with diode array detection using a YMC C30 3 µm 2.0 mm × 150 mm column.

Pro-Vitamin A carotenoid concentration (β-carotene equivalents)⁷: was calculated by sum of all-E-β-carotene + 1/2(β-cryptoxanthin) + α-carotene + Z-β-carotene isomers (9, 13, 15)).

Relative bioaccessibility (Micellarization efficiency)⁷:

$$\left(\frac{\text{Concentration in aqueous micellar fraction (AQ)}}{\text{Concentration in intestinal digesta (DG)}} \right) \times 100\%$$

Results

Stability of β-Carotene Equivalents during Processing (μg/g DW)						Micellarization Efficiency (%)					
#	Cultivar	Raw	Fermentation	Oven-Drying	Porridges	β-CRP	15-Z-β-C	13-Z-β-C	α-C	All-E-β-C	9-Z-β-C
1	GM 4414-5	31.1 ± 2.4	<div>28.0 ± 0.4</div>	15.8 ± 0.4	3.3 ± 1.6	17.6 ± 3.9	14 ± 3.9	12.4 ± 3.9	15.3 ± 4.7	10 ± 2.9	11.4 ± 3.5
				24.1 ± 0.4	17.2 ± 1.2	21.3 ± 1.7	9.1 ± 0.2	8.1 ± 0.3	6.5 ± 6.5	5.6 ± 0.3	7.8 ± 0.1
2	GM 4571-3	23.1 ± 0.7	<div>19.4 ± 1.7</div>	15.2 ± 0.4	9.7 ± 0.7	ND	14.9 ± 2.7	14.7 ± 1.9	ND	9.4 ± 1.8	13.4 ± 1.9
				5.4 ± 0.0	2.3 ± 0.1	ND	13.9 ± 2.8	12.9 ± 2.1	ND	10.1 ± 2.2	12.7 ± 2.2
3	GM 5194-5	26.0 ± 0.5	<div>18.8 ± 1.0</div>	14.5 ± 0.6	10.2 ± 0.0	ND	11.4 ± 5.7	8.7 ± 5.9	19.7 ± 0.7	8.7 ± 3.2	10.2 ± 3.3
				7.9 ± 0.1	4.6 ± 0.2	ND	11.9 ± 2.2	11.5 ± 1.6	ND	7.6 ± 0.7	11.7 ± 1.1
4	GM 5194-13	27.7 ± 0.3	<div>22.3 ± 0.2</div>	15.8 ± 0.4	9.5 ± 0.3	ND	12.3 ± 0.2	10.2 ± 0.2	ND	7.6 ± 0.2	9 ± 0.2
				5.1 ± 0.1	3.8 ± 0.1	18.6 ± 0.7	10.1 ± 1.6	9.8 ± 1.2	14.8 ± 1.5	6.5 ± 1	9.8 ± 1.3
5	GM 5212-6	42.7 ± 0.4	<div>34.2 ± 1.7</div>	27.3 ± 0.4	21.1 ± 0.7	15 ± 1.6	6.6 ± 1.4	5.6 ± 1.6	10.3 ± 3.4	3.7 ± 1	4.8 ± 1.2
				25.6 ± 1.0	18.0 ± 0.2	18.8 ± 1.9	11.9 ± 2	10 ± 1.6	13.6 ± 1.6	8.7 ± 2	10.5 ± 2.1
6	SM 3757-75	23.6 ± 1.0	<div>18.0 ± 0.6</div>	8.2 ± 0.2	6.0 ± 0.3	ND	20.6 ± 5.9	16.7 ± 4.6	17.7 ± 4.2	16.5 ± 4.8	18 ± 5.1
				5.6 ± 1.1	2.5 ± 0.1	26.7 ± 2.7	21.9 ± 4.5	20.4 ± 3.2	24.9 ± 4.8	16.5 ± 4	19.1 ± 4.6
7	SM 3758-43	27.1 ± 0.3	<div>26.2 ± 0.3</div>	15.1 ± 0.0	8.7 ± 2.9	ND	9.1 ± 0.8	8.1 ± 0.4	13 ± 2	4.3 ± 0.3	8.4 ± 0.8
				19.8 ± 0.5	13.6 ± 0.3	ND	9.8 ± 0.1	9.7 ± 1.1	ND	4.4 ± 1	8.9 ± 0.3
8	SM 3774-21	28.1 ± 2.6	<div>21.8 ± 1.7</div>	17.5 ± 0.1	10.1 ± 0.7	19.1 ± 1.8	12.7 ± 2.8	13.9 ± 2.5	18.3 ± 1.2	8.6 ± 1.9	13.5 ± 2.5
				12.4 ± 0.0	8.3 ± 0.0	18.9 ± 2	15.6 ± 2.5	13.7 ± 2	13.5 ± 1.9	13.7 ± 2.5	14.3 ± 2.6
9	SM 3762-15	24.9 ± 0.1	<div>23.9 ± 0.4</div>	16.1 ± 0.0	11.9 ± 0.6	14.1 ± 1.2	9.8 ± 2.4	8.6 ± 2.4	13.7 ± 1.3	5.7 ± 2.1	8.3 ± 2.4
				16.1 ± 0.2	18.6 ± 0.8	ND	16 ± 2.1	20.2 ± 5.3	23.7 ± 3.3	12.8 ± 1.6	17.3 ± 1.7
				12.3 ± 0.1	9.1 ± 0.4	22.9 ± 0.3	9.6 ± 1.4	9.9 ± 1.6	19.5 ± 1.9	5.3 ± 0.8	12.4 ± 2
10	SM 3767-84	28.48 ± 3.45	<div>21.53 ± 0.38</div>	13.5 ± 0.2	6.9 ± 0.5	ND	13.9 ± 2.7	11.6 ± 2.6	16.6 ± 1.8	9.2 ± 2.4	13.2 ± 2.2
				14.9 ± 0.4	13.0 ± 1.7	17.5 ± 2.9	10 ± 1.2	8.9 ± 0.8	15.5 ± 3.1	6.8 ± 1.0	8.6 ± 1.3
				5.3 ± 0.2	2.9 ± 0.1	13.3 ± 2.6	12.1 ± 0.8	11.2 ± 1.1	12.6 ± 1.3	9 ± 1.0	11.1 ± 1.4

Figure 4. Stability of β-carotene equivalents during processing (µg/g DW) and micellarization efficiency (%). Data represent mean ± SEM for 3 replicate of cassava extractions detected by HPLC with values representing sum of all carotene species with pro-vitamin A activity. Different color in the oven-drying column means that those samples were toasting (Gari) instead of oven-dried. C=Carotene, CRP=Cryptoxanthin. ND=No detectable.

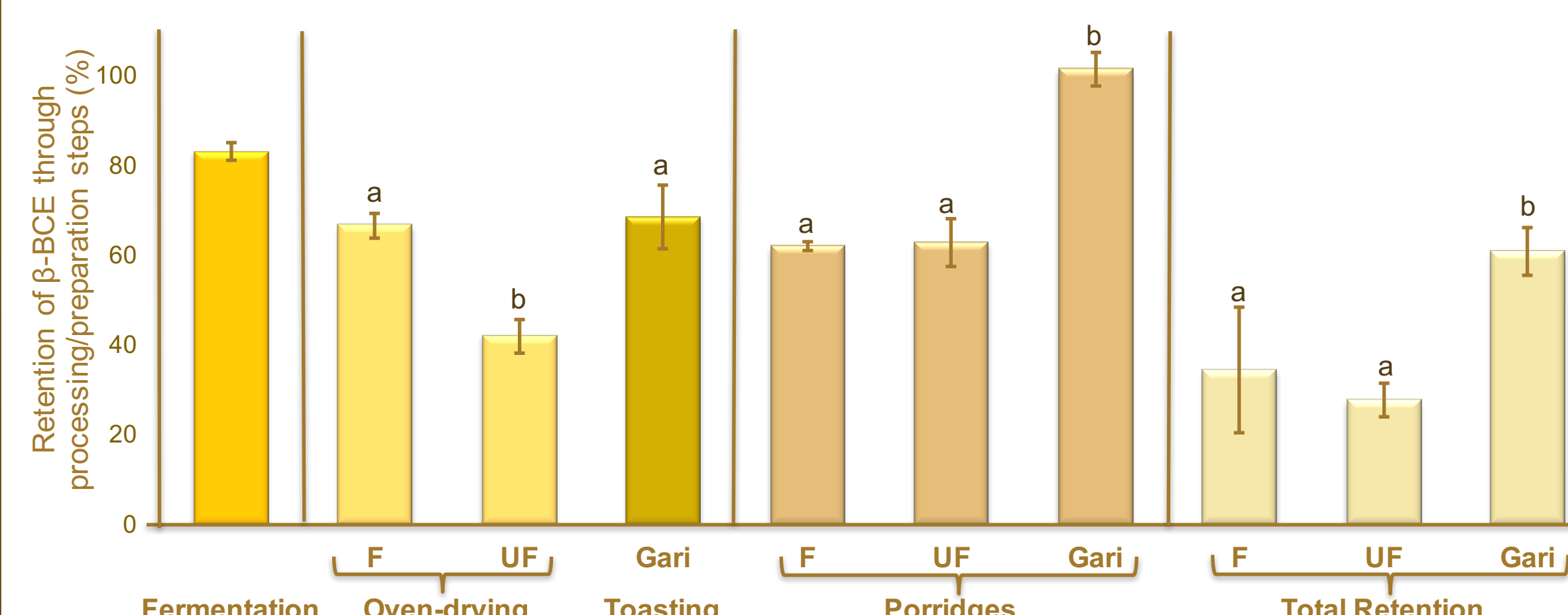


Figure 5. Retention of β-carotene equivalents through processing steps of raw cassava roots. Data represent mean ± SEM for 10 cassava cultivars. Different letters represent statistically significant differences (p<0.05). F= Fermented, UF= Unfermented.

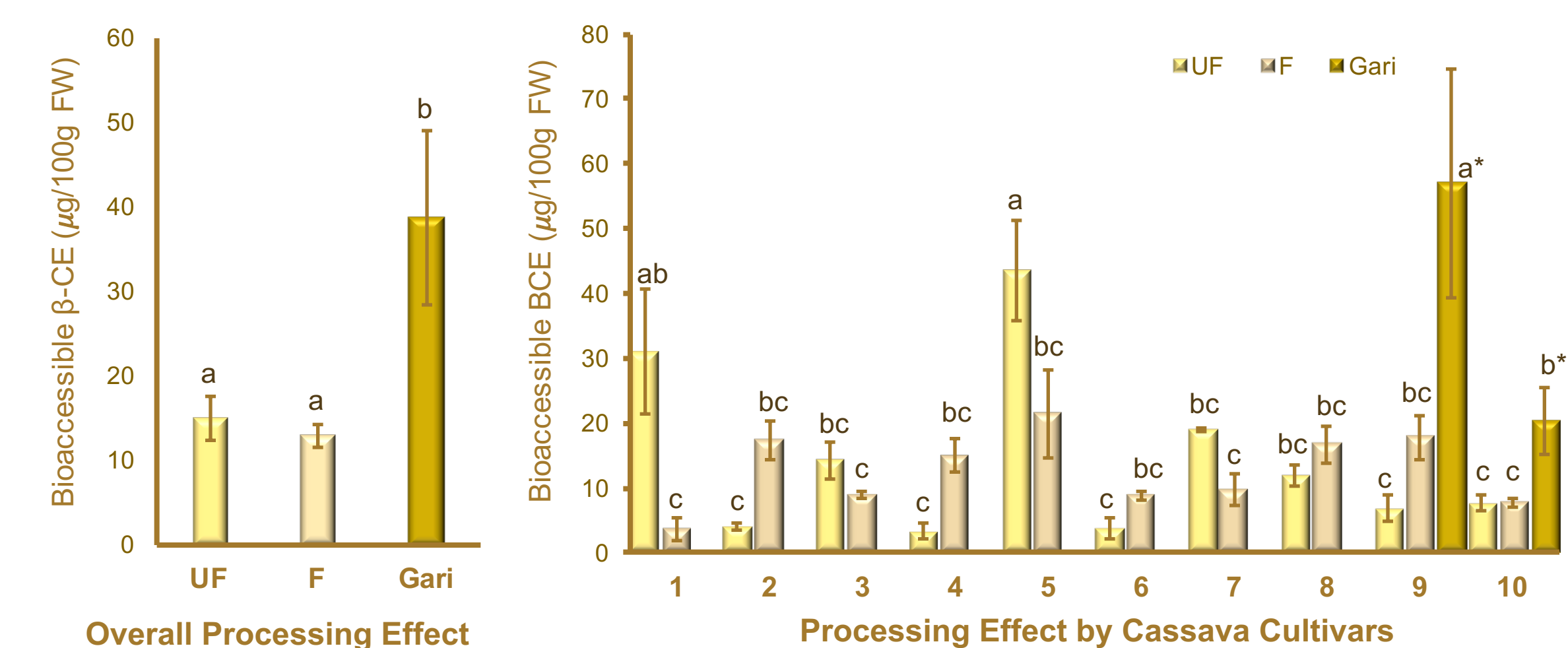


Figure 6. Bioaccessible β-carotene equivalents (µg/100g FW) from cassava porridges. Data represent mean ± SEM for unfermented (UF), fermented (F) and Gari porridges from 10 cassava cultivars. Different letters represent statistically significant differences (p<0.05). F= Fermented, UF= Unfermented.

Conclusions

- Around 80% of the β-carotene equivalents are retained during fermentation of grated biofortified cassava roots.
- Fermented cassava showed higher β-carotene equivalents retention after oven-drying (p=0.007) in comparison to unfermented cassava. However, no significant difference (p>0.05) was observed between oven-drying and toasting of fermented cassava roots.
- No significant difference (p>0.05) in the total β-carotene equivalent retention was observed between fermented and unfermented biofortified cassava roots when they were processed into porridges. Gari processing however, seems to promote higher retention and bioaccessibility of β-carotene equivalents.
- Selected cassava cultivars showed higher bioaccessibility of pro-vitamin A carotenoids following the fermentation process, suggesting that genotypic and/or another factors merit further investigation for their role in facilitating carotenoid stability and bioaccessibility from biofortified cassava products.

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